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Physiological Evaluation of Groundnut (*Arachis hypogaea* L.) Varieties for Salt Tolerance and Amelioration for Salt Stress

Nithila S., Durga Devi D., Velu G., Amutha R. and Rangaraju G.
Department of Crop Physiology, Tamil Nadu Agricultural University, Coimbatore - 641 003, INDIA

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Abstract

Soil salinity and sodicity cause detrimental effects on plant activities, which are likely to alter the yielding potential of the crops. Hence to identify the physiological parameters, which get altered under salt stress conditions and measuring the quantum of damage caused by the stress is the need of the hour. To standardize the method of ameliorating the adverse effects of sodicity, the present investigations were carried out in groundnut under three conditions viz., laboratory, pot culture and field. In the laboratory screening study, ten varieties were subjected to three levels of salinity stress, viz., 50 mM, 100 mM, 125 mM NaCl and three levels of sodicity stress viz., 25 mM, 50 mM and 75 mM $NaHCO_3$. Based on mean stress tolerance index (STI) TMV7, CO5, JL24 and BSR1 recorded lesser STI of 13 to 19 under high salinity and 15 to 24 under high sodicity levels. Therefore, by rejecting these four varieties, the other six varieties were further evaluated under pot culture condition were subjected to two levels of salinity stress (50 mM and 100 mM NaCl) and two levels of sodicity stress (25 mM and 50 mM $NaHCO_3$). The groundnut variety CO4, identified as tolerant variety and ALR3, as susceptible variety, through pot culture experiment. To assess the influence of different plant growth regulating chemicals on alleviating the adverse effects of sodicity stress, field study was conducted using CO4 and ALR3 under sodic soil condition. Brassinolide 1 ppm sprayed at pre flowering, pegging and pod formation stages was highly effective in overcoming the adverse effects of salt stress through enhancing the overall physiological efficiency of the crop and in improving the pod yield even in the varieties sensitive to salt stress.

Keywords: Salinity, sodicity, ground nut, leaf area, proline, catalase, brassinolide and pod yield.

Introduction

A salt affected soil is defined as one that has been adversely affected to the extent that it is no longer suitable for the growth of most crops by the presence of action of soluble salts. This group of soils includes both saline and sodic soils. Soil salinity and sodicity problems are present in nearly every irrigated area of the world and also occur on non irrigated croplands. Thus, virtually no land is immune from salinization. Therefore, for sustaining life on earth, control of these problems and finding new ways to utilize these extensive saline and sodic soils and water resources, at least for agricultural purposes, are vital and urgent. Reclamation, or at least minimizing the effect of salinity and or sodicity, is important and necessary. Other ways to develop ability of plants to survive and maintain their growth under saline conditions is known as salt tolerance. There is a continuous spectrum of plant tolerance to saline conditions ranging from glycophytes that are sensitive to salt, to halophytes, which survive in very high concentrations of salt[1]. Groundnut is an important commodity in many developing countries, particularly in India where the nitrogen rich crop residues are also used as fodder. The production of groundnut in India needs to be increased from the current 8 million to about 14 million tonnes by 2020 to meet the increasing demand of the oil and confectionery industries. This increase will have to be practically achieved by growing groundnut in lands considered so far as unsuitable for agriculture, including salt affected soils. This may lead to find gene source as well as methods for screening large number of genotypes for salt tolerance. A better understanding of the mechanisms, by which, plants respond to salinity / sodicity stress may help in developing more tolerant varieties. Besides this, development of amelioration technology may pave the way for improving growth and yield of crop plants, particularly sensitive species, growing under the hostile environments. Based on these backgrounds, the present study was formulated.

Materials and Methods

The lab and pot culture experiment was conducted at department of crop physiology and Anbil Dharmalingam Agricultural college and Research Institute, Tamil Nadu Agricultural University, Coimbatore to screen the varieties of groundnut for salinity and sodicity tolerance and to understand the physiological and biochemical mechanisms of tolerance to salinity and sodicity stresses. An attempt was also made to alleviate the sodicity stress with foliar spray of plant growth regulating chemicals and nutrients. In the laboratory screening study, ten varieties viz., CO1, CO2, CO3, CO4, TMV2, TMV7, ALR3, VRI 2, JL24 and BSR1 were subjected to three levels of salinity stress, viz., 50 mM, 100 mM, 125 mM NaCl and three levels of sodicity stress viz., 25 mM, 50 mM and 75 mM

$NaHCO_3$. Based on mean stress tolerance index (STI) TMV7, CO5, JL24 and BSR1 recorded lesser STI of 13 to 19 under high salinity and 15 to 24 under high sodicity levels. Therefore, by rejecting these four varieties, the other six varieties were further evaluated under pot culture condition for their morpho-physiological characters besides yield were subjected to two levels of salinity stress (50 mM and 100 mM NaCl) and two levels of sodicity stress (25 mM and 50 mM $NaHCO_3$). The groundnut variety CO4, identified as tolerant variety and ALR3, as susceptible variety, through pot culture experiment. To assess the influence of different plant growth regulating chemicals on alleviating the adverse effects of sodicity stress, field study was conducted using CO4 and ALR3 under sodic soil condition in Anbil Dharmalingam Agricultural College and Research Institute, Tamil Nadu Agricultural University, Trichy.The experiment was laid out in a Split plot design replicated four times. There were nine treatments viz., control, $CaCl_2$ 1 %, BR 0.5 ppm, BR 1 ppm, SA 50 ppm, SA 100 ppm, KNO_3 1 %, DAP 2 %, Nutrient mixture {DAP (1 %) + KNO_3 (0.5 %) + $FeSO_4$ (0.5 %) + Borax (0.2 %) + NAA (20 ppm) + SA (50 ppm) + BR (1 ppm)}. These treatments were imposed as foliar sprays on 25th, 55th, and 85th DAS coinciding with preflowering, pegging and pod formation stages. Stress tolerance index was calculated using the formula proposed by Dhopte and Livera and expressed as per cent[2]. Proline content of the leaf was estimated by the method of Bates *et al.,* and expressed as µg g^{-1} fresh weight [3]. Catalase activity was assayed as per the procedure adopted by Gopalachari and expressed as µg H_2O_2 /g/min[4] .

Results and Discussion

Laboratory Experiment: Stress tolerance indices of these varieties under severe salinity and sodicity stresses were very much low with the range of 13 to 19 (salinity) and 15 to 25 (sodicity) per cent. The highest stress tolerance index of 64 was observed in CO4 under severe salinity stress, whereas CO2 showed highest tolerant index of 60 under sodicity condition (figure 1 and 1a). The performance of other varieties such as VRI2, ALR3 and CO3 was also comparatively better under both severe salinity and sodicity stress conditions. Based on these observations made, the varieties viz., TMV2, CO5, JL24 and BSR1 were categories as highly sensitive and susceptible to both salinity and sodicity stresses and, therefore, these varieties are considered as unsuitable for cultivation in saline and sodic soils. Sharma *et al*., also conducted similar experiment and reported that some varieties of Greengram were more sensitive to NaCl, $CaCl_2$ and Na_2SO_4, while other to Na_2HCO_3 and (Na_2CO_3)[5].

Pot culture Experiment: Leaf area, the photosynthetic surface of the plant, was drastically reduced under salinity and sodicity stresses. High level of salinity stress caused a mean leaf area reduction of 8.3 per cent, whereas sodicity at higher level resulted in 10.7 per cent reduction. CO4 followed by VRI2 performed better by maintaining significantly higher leaf area with lesser reduction in salinity and sodicity stresses. ALR3, however, showed drastic reduction by 19 per cent and 15 per cent over control under sodicity and salinity stresses respectively (table 1). These results were supported by Hooda *et al*., who reported that plants that were subjected to salt stress showed a greater reduction in leaf area[6].

The salinity and sodicity stresses induced proline accumulation at various levels. At the time of pegging, salinity and sodicity at lower levels caused an increase in proline accumulation by 15 and 16 per cent respectively over control. Higher levels salinity and sodicity stresses, however, resulted in 8 and 9 per cent increase over control (table 2). Therefore it was revealed that proline synthesis mighty have accelerated at the sub lethal level of stress rather than severe stress. These results are strongly supported by Muthukumarasamy and Panneerselvam who reported that NaCl salinity induced the accumulation of proline in all parts of peanut seedlings with increased accumulation at lower NaCl level[7]. Girija *et al.*, also observed similar results in various groundnut genotypes[8].

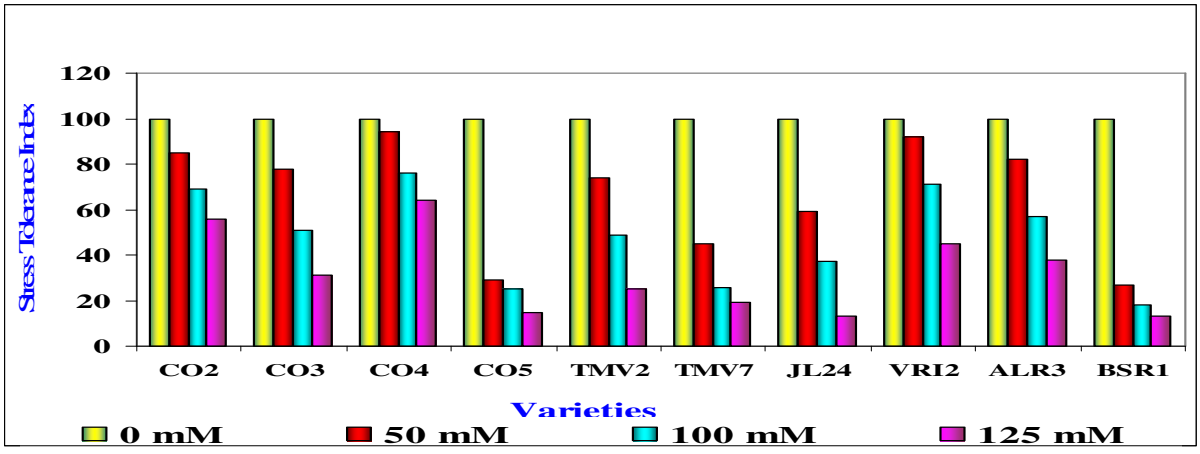


Figure-1
Effect of salinity on Stress Tolerance Index of groundnut seedlings

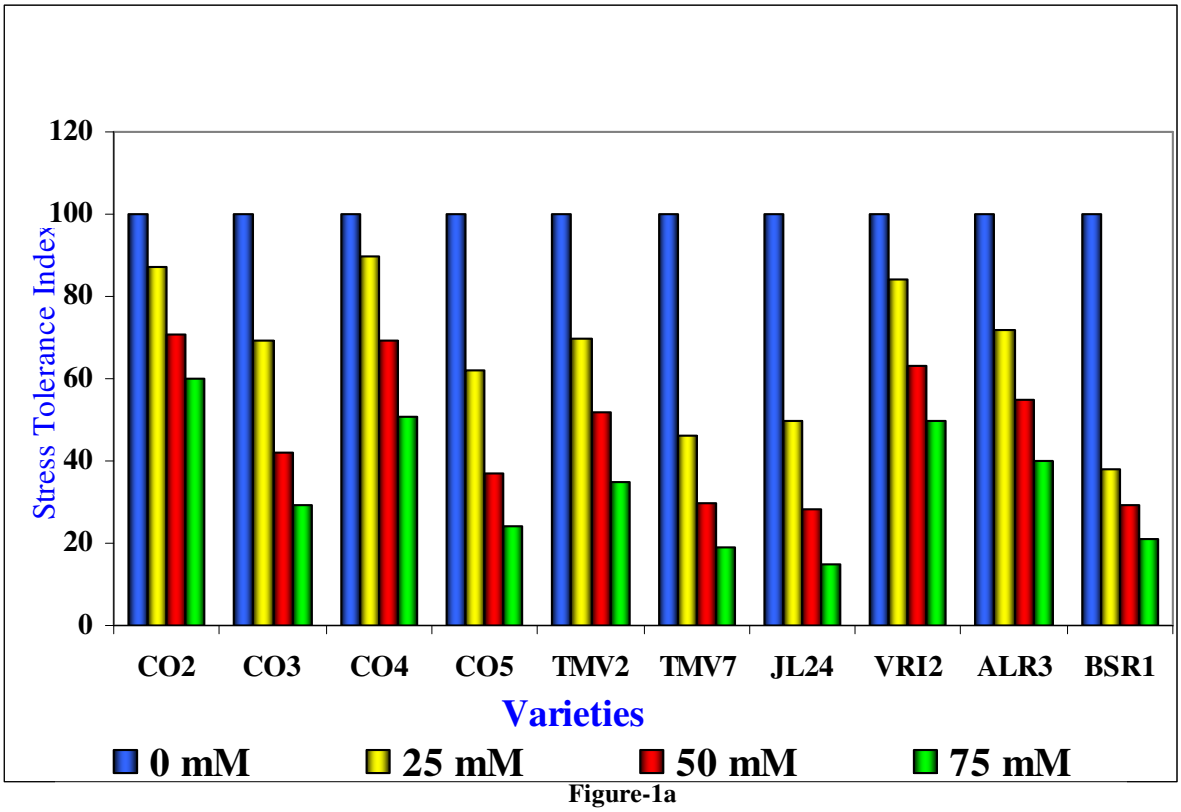


Figure-1a
Effect of sodicity on Stress Tolerance Index of groundnut seedlings

Table-1
Effect of salt stress on leaf area (cm^2 $plant^{-1}$) of groundnut varieties at different growth stages

Treatments	25 DAS	40 DAS	55 DAS	70 DAS	85 DAS	Mean
Factor I						
V_1	247.7	477.7	682.2	988.8	567.5	**592.8**
V_2	223.5	398.3	528.3	905.1	452.1	**501.5**
V_3	281.0	659.0	811.8	1024.7	678.5	**691.0**
V_4	265.0	511.7	702.0	1001.7	619.6	**620.0**
V_5	223.1	460.1	651.2	937.1	534.5	**561.2**
V_6	178.7	375.4	498.3	797.1	425.0	**454.9**
Mean	**236.5**	**480.4**	**645.6**	**942.4**	**546.2**	**570.2**
SEd	**1.77**	**3.65**	**4.79**	**6.91**	**4.06**	
CD((P=0.05)	**3.54**	**7.31**	**9.59**	**13.83**	**8.13**	
Factor II						
T_1	303.0	607.3	717.7	992.6	623.2	**648.8**
T_2	241.8	544.4	666.4	969.4	582.4	**600.9**
T_3	220.7	373.5	618.5	910.2	513.5	**527.3**
T_4	224.6	522.1	651.9	953.5	553.3	**581.1**
T_5	192.3	354.5	573.6	886.4	458.5	**493.1**
Mean	**236.5**	**480.4**	**645.6**	**942.4**	**546.2**	**570.2**
SEd	**1.62**	**3.34**	**4.38**	**6.31**	**3.71**	
CD(P=0.05)	**3.23**	**6.67**	**8.75**	**12.62**	**7.42**	

Table-2
Effect of salt stress on proline content (µg g^{-1}) of groundnut varieties at different growth stages

Treatments	25 DAS	40 DAS	55 DAS	70 DAS	85 DAS	Mean
Factor I						
V_1	246	361	507	393	260	**356**
V_2	255	325	471	350	194	**321**
V_3	323	398	574	440	284	**408**
V_4	308	376	541	410	271	**380**
V_5	270	345	488	375	219	**343**
V_6	258	307	464	279	157	**294**
Mean	**277**	**352**	**508**	**374**	**231**	**350**
SEd	**2.1**	**2.6**	**3.8**	**2.8**	**1.7**	
CD(P=0.05)	**4.1**	**5.2**	**7.6**	**5.6**	**3.5**	
Factor II						
T_1	208	312	464	323	196	**302**
T_2	309	373	533	389	246	**371**
T_3	264	334	500	343	214	**330**
T_4	320	385	538	440	268	**396**
T_5	284	356	504	376	230	**352**
Mean	**277**	**352**	**508**	**374**	**231**	**350**
SEd	**1.9**	**2.4**	**3.5**	**2.5**	**1.6**	
CD(P=0.05)	**3.8**	**4.7**	**6.9**	**5.1**	**3.2**	

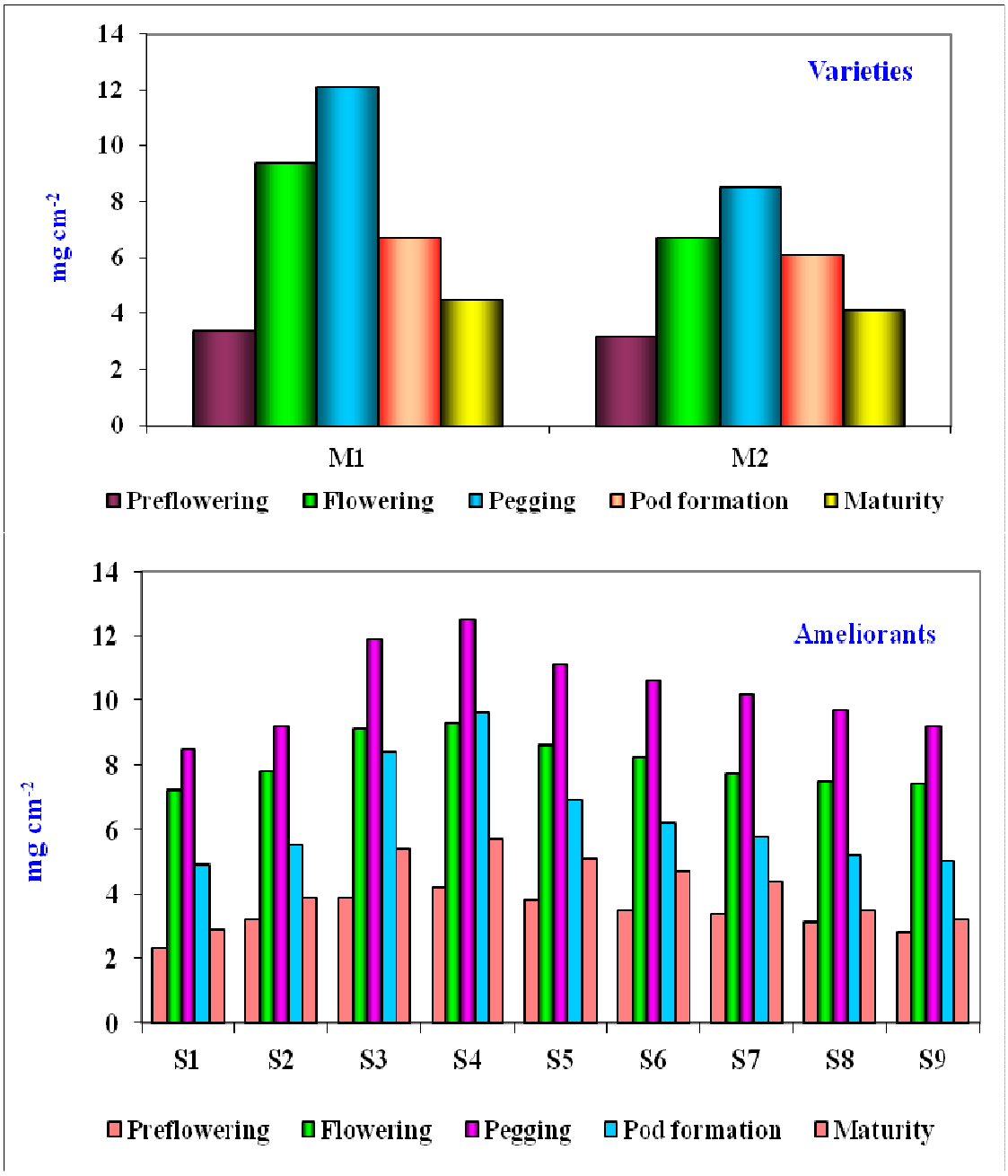
Field Experiment: The influence of plant growth regulators and nutrients sprayed at preflowering, pegging and pod formation stages on the two groundnut varieties, CO4 and ALR3 exhibited significant variations at all phenological stages.

As observed in the present study, CO4 recorded the highest Specific Leaf Weight of 14.8 mg at pegging and maintained higher levels at the subsequent stages (figure 2). Most of the ameliorative chemicals showed positive effect on SLW and BR 1 ppm with maximum effect in both the varieties. As per the report of Lugg and Sindair, the maximum SLW, as influenced by brassinolide, was highly correlated with leaf photosynthesis in several crops[9].

The effect of plant growth regulating chemicals on controlling the transpiration rate could be observed from the present study in both the groundnut varieties. Brassinolide and salicylic acid were effective in lowering the rate of transpiration through enhancing the stomatal diffusive resistance. Greater than 40 per cent increase in SDR caused about 20 per cent reduction in transpiration rate due to the application of BR 1 ppm in both the varieties (figure 3). Yeo and Flowers suggested that high diffusive resistance of stomata coupled with low transpiration rate led to the less accumulation of toxic ions such as Na^+ and Cl^- in shoot, favors better growth in rice [10].Similar results were observed in Sorghum cultivar[11].

Saha and Gupta recorded a low rate of catalase activity under salinity stress and however the activity could be enhanced by the application of growth regulating chemicals[12].Similar results were observed in barley crop.[13] The two groundnut varieties employed in the present study showed differential responses to the stress ameliorative chemicals. CO4 though maintained higher activity than ALR3, the response to growth regulating chemicals was more in ALR3. In both the varieties BR and salicylic acid were found effective in enhancing the enzymatic activity remarkably (table 3).

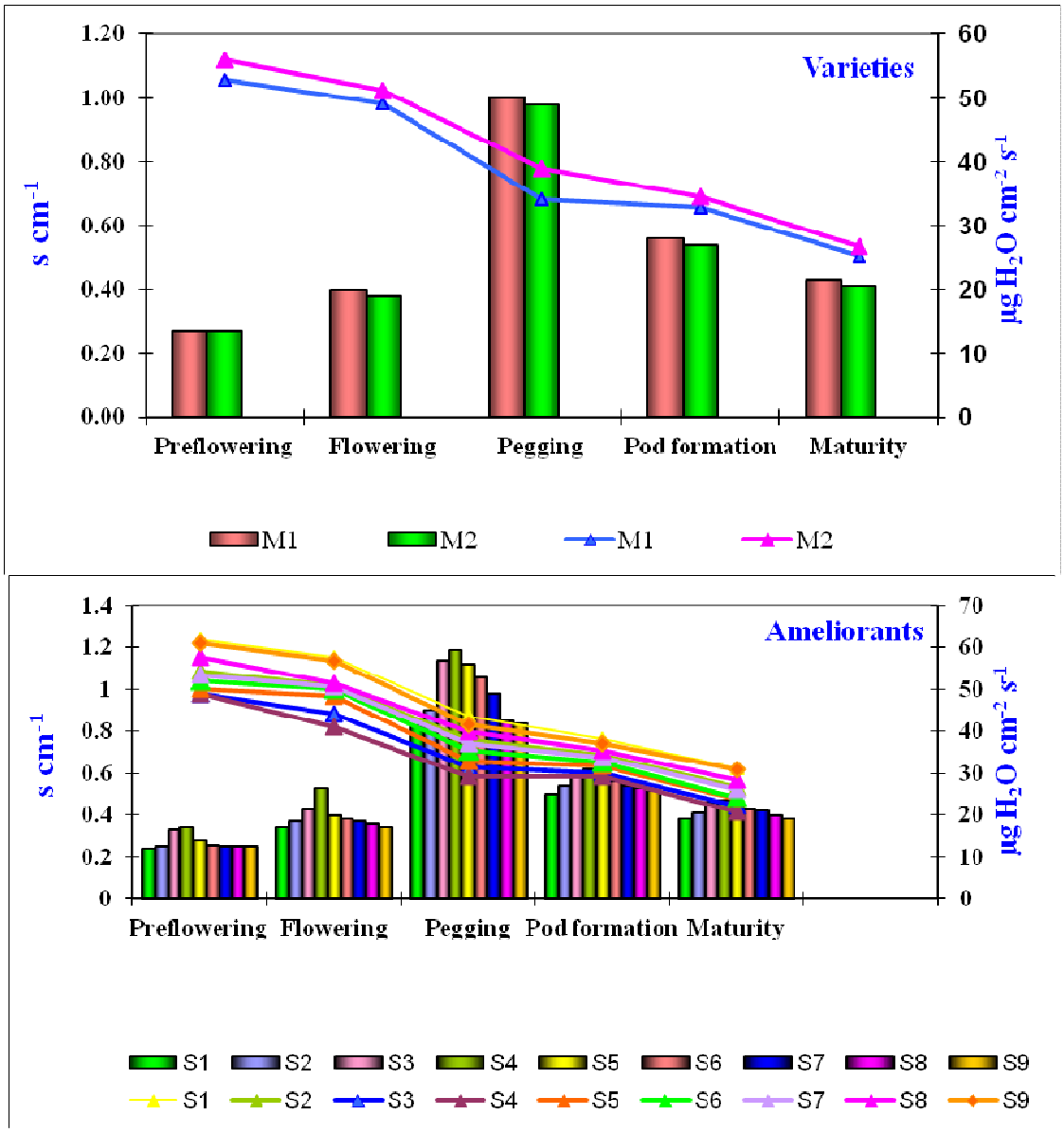
Pod yield of groundnut, contributed by number of pods and pod size, greatly affected by sodicity. ALR3 showed greater reduction than CO4 (Table 3). As reported by Vidyavardhini and Rao, this inhibiting effect of salinity stress could however be reversed by application brassinolide in the form of 24 epibrassinolide or 28 homobrassinolide[14]. They also further stated that BR not only removed the inhibitory effect of salinity, but also promoted the growth and yield of crops. In the present study also, the beneficial role of BR could be revealed through yield improvement of two groundnut varieties under sodicity condition. CO4 registered a 15 per cent yield increase, whereas ALR3 showed 13 per cent yield increase over control due to the application of BR at 1 ppm concentration. According to them BR is assured to increase the sink capacity by promoting translocation and accumulation of starch within the reproductive parts, resulting in promotion of maturation of the kernels.



M_1 – CO4 **M_2 – ALR3**

S_1	-	Control	S_6	-	SA 100 ppm
S_2	-	$CaCl_2$ 1 %	S_7	-	KNO_3 1 %
S_3	-	BR 0.5 ppm	S_8	-	DAP 2 %
S_4	-	BR 1 ppm	S_9	-	Nutrient mixture
S_5	-	SA 50 ppm			

Figure-2
Effect of ameliorative chemicals on specific leaf weight at different growth stages of groundnut varieties



M_1 – CO4 M_2 – ALR3

S_1 - Control
S_2 - $CaCl_2$ 1 %
S_3 - BR 0.5 ppm
S_4 - BR 1 ppm
S_5 - SA 50 ppm
S_6 - SA 100 ppm
S_7 - KNO_3 1 %
S_8 - DAP 2 %
S_9 - Nutrient mixture

Figure-3
Effect of ameliorative chemicals on transpiration rate and stomatal diffusive resistance at different growth stages of groundnut varieties

Table-3
Effect of ameliorative chemicals on catalase activity (μg H_2O_2 min^{-1} g^{-1}) and pod yield(kg ha^{-1}) of Groundnut varieties

Treatments	Pre flowering	Flowering	Pegging	Pod formation	Maturity	Mean	Pod yield (kg ha^{-1})
Main plot							
M_1	61.16	71.92	83.80	62.40	49.34	**65.73**	1702
M_2	47.73	62.15	72.05	54.04	37.96	**54.78**	1590
Mean	**54.44**	**67.04**	**77.92**	**58.22**	**43.65**	**60.25**	**1646**
SEd	**0.042**	**0.033**	**0.019**	**0.013**	**0.016**		**0.1**
CD(P= 0.05)	**0.080**	**0.040**	**0.080**	**0.058**	**0.032**		**0.7**
Sub plot							
S_1	41.09	56.79	65.53	39.46	28.45	**46.26**	1538
S_2	49.91	63.49	74.55	56.72	41.04	**57.14**	1608
S_3	63.45	73.70	84.94	64.79	52.17	**67.81**	1725
S_4	67.95	79.64	93.40	77.80	57.89	**75.33**	1754
S_5	64.38	74.99	86.84	73.06	52.44	**70.34**	1679
S_6	59.37	68.32	81.08	61.33	47.79	**63.58**	1658
S_7	52.94	64.65	76.89	58.28	44.64	**59.48**	1640
S_8	46.48	61.93	70.33	48.33	37.88	**52.99**	1621
S_9	44.43	59.83	67.77	44.24	30.56	**49.36**	1593
Mean	**54.44**	**67.04**	**77.92**	**58.22**	**43.65**	**60.25**	**1646**
SEd	**0.672**	**0.813**	**0.946**	**0.721**	**0.633**		**9.9**
CD(P=0.05)	**1.369**	**1.656**	**1.927**	**1.468**	**1.265**		**20.1**

Conclusion

From the studies, it is concluded that the groundnut variety CO4 was identified as the most tolerant variety to salt stress and ALR3, the most sensitive one. Maintenance of optimum leaf area with high proline, transpiration rate and stomatal diffusive resistance were the physiological basis for tolerance to both salinity and sodicity stresses. Brassinolide 1 ppm sprayed at preflowering, pegging and pod formation stages was highly effective in overcoming the adverse effects of salt stress through enhancing the overall physiological efficiency of the crop and in improving the pod yield even in the varieties sensitive to salt stress.

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